REMARKS

Claims 1-19 are pending; claims 17-19 have been withdrawn from consideration; claims 1-16 are rejected.

Upon entry of this amendment, claims 12 and 17-19 will be canceled, and claims 1-11 and 13-16 will be pending.

Claims 17-19 have been cancelled in view of the Restriction Requirement. Applicants reserve the right to file the cancelled claims in a later divisional application.

The remainder of the claims have been amended to more clearly recite that which Applicants regard as their invention, and to more fully comply with U.S. format.

Applicants include herewith a substitute Specification, in both "marked up" and "clean" forms, correcting grammatical errors and removing hyperlinks.

No new matter has been added. Entry of the amendment is respectfully requested.

I. Sequence Listing

At page 2, paragraph 2, of the Office Action, the Examiner notes that the each sequence disclosed in the specification is not referenced by a "SEQ ID number."

In response, Applicants include herewith a substitute specification, inserting the sequence identifiers pointed out by the Examiner, and correcting the grammatical errors and misspellings also noted by the Examiner. For the Examiner's convenience, Applicants also enclose a "marked-up" version of the substitute specification so the Examiner may easily see where the changes have been made. Applicants respectfully request entry of the substitute specification.

The Examiner also requires the submission of a new computer readable form, and paper copy, of the sequence listing. Applicants respectfully note that the Examiner has not indicated

that any errors exist in the Sequence Listing filed in this application on May 18, 2001, or if the Sequence Listing filed on May 18, 2001, has been misplaced. However, to further prosecution of this application in a timely fashion, Applicants are submitting herewith a revised Sequence Listing (and corresponding Statement), and request entry of the same as the Sequence Listing for this application.

II. Objections to the Specification

A. At page 2 of the Office Action, last paragraph, the Examiner states that the embedded hyperlinks, such as at page 2, line 4, must be deleted from the specification.

In response, Applicants note that each of the hyperlinks has been deleted from the substitute specification filed herewith.

Accordingly, Applicants respectfully request reconsideration and withdrawal of this objection.

B. At page 3 of the Office Action, first paragraph, the Examiner states that there are a number of informalities and misspellings in the specification that require correction.

In response, Applicants assert that each of the informalities and misspellings in the specification has been corrected in the substitute specification filed herewith.

Accordingly, Applicants respectfully request reconsideration and withdrawal of this objection.

III. Objections to the Claims

At pages 3 and 4 of the Office Action, the Examiner objects to many of the claims due to informalities and misspellings.

In response, Applicants include herewith an amendment to the claims, correcting each of the informalities and misspellings.

Accordingly, Applicants respectfully request reconsideration and withdrawal of this objection.

IV. Rejection of the Claims Under 35 U.S.C. §101

At page 5 of the Office Action, last paragraph, claims 1-16 are rejected under 35 U.S.C. §101 as lacking a patentable utility.

The Examiner states that the claimed invention is not supported by a specific, substantial and credible utility, or a well-established utility.

The Examiner explains that the method does not serve to identify anti-infective candidates. She also states that the method is not supported by a substantial utility because no substantial utility has been established for the claimed subject matter. The Examiner further states that the skilled artisan would have reason to doubt that sequence similarity alone would reasonably support the assertion that the biological activity of the claimed subject matter would be the same as that of the similar sequence.

In response, Applicants respectfully note that the Examiner appears to be interpreting the claims as a method for identifying proteins that can be used as anti-infectives, rather than what is actually being claimed, i.e., a method for identifying candidate proteins that may be useful as anti-infectives. Thus, rather than claiming a method of identifying proteins with a particular function, the method is used to identify a group of proteins that may have a particular function. Because of the large number of proteins that a researcher would need to screen in order to

identify anti-infectives, the claimed method has a clear utility in narrowing that group of proteins to a manageable number that can be screened through traditional biochemical analyses.

Thus, Applicants assert that the instant application recites a method for identifying candidate proteins that has a specific, substantial and credible utility.

Specifically, the claims are specific to a method of identifying candidate proteins of pathogens for use as anti-infectives. The method is specific to proteins that may be used as anti-infectives, as opposed to being a general utility that may be used to identify any of a number of other classes of proteins. By performing step vi) of claim 1, protein sequences are validated as candidate anti-infectives by comparing them to protein sequences of known proteins that have previously been biochemically characterized.

Thus, the method narrows the extremely large number of proteins produced by any given microorganism to a small group of candidate proteins. Therefore, the utility is specific to the subject matter claimed.

Second, the method recited in the present application is "substantial." It is a real-world use for identifying candidate proteins for further testing. The method is advantageous in that it allows a researcher to significantly narrow the scope of proteins that must be further biochemically characterized. In contrast to the examples given in the patent examiner training materials for determining utility, the present application is not claiming the proteins identified by the claimed method, but instead is claiming a method of identifying candidate proteins for further study.

No further research would need to be conducted to identify the candidate proteins for which the claims recite a method of identifying. This would be in contrast to claims to the

proteins themselves. In the latter case, additional experiments would need to be performed to determine whether the claimed proteins have a utility, e.g., use as an anti-infective.

Thus, in contrast to the Examiner's position, a substantial utility has been established for the claimed subject matter, i.e., a method of identifying candidate proteins, not a method of identifying proteins that will function as anti-infectives as the Examiner appears to interpret the claims.

Finally, it is clear that the claimed method of the present invention is credible. The skilled artisan reviewing the claims and specification would find the claimed method to be believable and credible for use in its recited purpose.

Applicants include the following comments as a means of providing additional explanation for the utility of the invention.

The development of any new or novel chemotherapeutic drug, vaccine or diagnostic kit for treatment of a disease may be based on information determined from the sequences of proteins isolated from a microbe causing the disease. In other words, the creation of a new or novel chemotherapeutic drug, vaccine or diagnostic kit may rely on the structural and functional attributes of a microbial protein that functions as an antigen and/or behaves as a toxin. This allows the production of a new or novel chemotherapeutic drug, vaccine or diagnostic kit having antagonistic attributes against the microbial antigen. However, the characterization of all protein sequences from a given microbe, by conventional means which comprise conducting sequence analyses, can be very time consuming and costly, and it may take from a few months to many years to identify and develop an efficient chemotherapeutic drug, vaccine or diagnostic kit for use in treatment of infections caused by a particular microbe.

With the identification of the genome sequences for many known pathogens, the approach for design and development of a chemotherapeutic drug, vaccine or diagnostic kit has become faster. This has been made possible by means of databases containing the above-mentioned protein sequences, and the application of bioinformatics to genomics and proteomics. The use of database information can function in manner similar to the search and identification of a book, an article, a published paper or a thesis in a library catalog or searching a file, a word or topic in MS word. In other words, bioinformatics of genomics and proteomics performs a similar function as the thesaurus, wherein there is complete compilation of all the synonyms and antonyms, i.e., the similar and different proteins sequences, but they are all anti-infective candidates clustered together and then one may choose and identify the right word for framing sentences, i.e., then choosing and identifying the right protein sequence of interest for drug design.

The instant invention is also based on similar lines wherein the designed database is a repository created for identification and analysis of protein sequences which are or can be potential candidates for development of efficient chemotherapeutic drugs, vaccines or diagnostic kits for disease. The non-obviousness to the person skilled in art lies with the fact the database was designed after careful analysis of only those potential anti-infective candidates, which are outlier proteins. These outlier proteins are either virulence proteins or antigens or used as drug targets. The claimed method detects outlier proteins using cluster analysis of quantified sequence attributes such as hydrophobicity, charge, amino acid composition and dipeptide complexity. The physico-chemical characteristics such as charge, hydrophobicity and compositional analysis are fundamental characteristics of a protein. The fact that the majority of

proteins fall into a large cluster indicates that majority of the proteins encoded in a genome belong to a set of basic physico-chemical characteristic specified in the organism. Out of this, a few proteins appear as outliers indicating that they are fundamentally different from the majority. These proteins thus have a likelihood of belong to a set of evolutionary processes for some specialized function that may be characteristic of the organism and its biology.

Amino acid compositional analysis of proteins, including residue pair frequencies inferred from available protein sequences, has lead to several predictive methods of wide interest. The classification of proteins into different groups of function and folding types (Nishikawa et al, 1983; Ramachandran et al, 1988), the folding type of a protein (Nakashima et al, 1986), the clustering of proteins using all possible pairs of amino acids as structural descriptors (Nakayama et al, 1988) were based on compositional characteristics. Further, the distinction of cytoplasmic and extracellular domains of transmembrane proteins (Nakashima et al, 1992), the discrimination of intracellular and extracellular proteins (Nakashima et al, 1994), the prediction of in vivo stability of a protein (Guruprasad et al, 1990) were all based on composition characteristics. Recently, the prediction of potentially secreted proteins in bacteria (Schneider et al, 1999) has been developed.

Although these data suggest that amino acid composition and residue pair frequencies are important attributes with which features such as sub-cellular location or protein stability can be correlated, other attributes such as hydrophobicity and charge also contribute to protein structure, stability, and function. Electrostatic charges confer stability to proteins and it has been observed earlier that organisms living in hyper-thermophilic environments have a high proportion of charged residues in their proteins (Natesh et al, 1999; Mande et al, 2000; Nandi et al, 2002).

Similarly, long hydrophobic regions (17-21 amino acids) are indicative of interactions with membrane structures. Information from hydrophobic scales has also been used to predict the residues likely to be buried in a protein. This has lead to the development of a predictive method for identifying residues that, when mutated, may lead to temperature sensitivity (Varadarajan et al, 1996). Thus a vast number of predictions have been made using the fundamental characteristics of proteins.

With reference to the work of Applicants, the proof of the concept lies in the fact that some of these outlier proteins, identified in several different organisms, have been already established as having anti-infective roles. These are indicated in the examples provided in the specification. The above mentioned principle, along with the proof of concept, constitutes a body of facts that can enable any experimenter to clone and produce proteins to be used as anti-infectives.

The method provided in the application has vastly simplified the traditional route that uses the "gene by gene" approach. Given the enormous information available but the lack of tools to process it in a mega-scale, the instant method provides a short list of candidates for the experimenter. The outlined method is inherently based on fundamental characteristic of proteins and well-established bioinformatics analysis. There are many such approaches that have been used by other investigators using bioinformatics (e.g., Chakravarti, D.N. et al: 2000a & 2000b) towards similar objectives.

The anti-infective proteins have the following features:

1. An anti-infective should be a protein against which reagents are developed such as inhibitors or antibodies to counter the attack of the invading pathogen.

2. The protein should be one such that the reagents rendering it ineffective do not cross-react with the human host. Thus, anti-infective proteins should be sufficiently different in fundamental characteristic from the host proteins. It is this aspect that was tested using standard bioinformatics tools for similarity and homology, and that was used in developing proteins for use as anti-ineffective by the inventors.

The principles on which the method of the instant application has been developed are fundamental. Other methods employed in the present invention are standard time-honored bioinformatics tools. A large body of work in biotechnology and medicine has been researched and published, and products produced based on these bioinformatics tools are used in this work. The evidence in the literature describing the use of these tools is overwhelming. Several references are cited below for the Examiner's convenience, both with general and specific relevance to the instant application.

Applicants also contend that the majority of the world accepts the principle that "Homologous sequences fold similarly and have the same function." A vast amount of bioinformatics analysis, such as those carried out by investigators like Koonin, Galperin, Gelfland, Bork and other colleagues, have primarily rested on sequence analysis using bioinformatics tools such as the BLAST family of algorithms, transmembrane predictions and so on. Their analyses, and the development of the COG database and related databases at EBI/EMBL and Argonne National Laboratories have all rested on the above-mentioned principle. These databases are used currently in many drug discovery strategies.

Even present day bioinformatics companies use these standard techniques for experimental work. Therefore, the information provided along with proof of concept constitutes a body of facts for the experimenter to use in the development of proteins as anti-infectives.

While there are multiple approaches to the identification and development of anti-infectives, the present method is a novel *in silico* method.

The application of bioinformatics through the use of genomics and proteomics can therefore expedite the development of efficient chemotherapeutic drugs, vaccines or diagnostic kits for disease by identifying potential candidates for such studies. The database also allows comparison and analysis of the already available protein sequences between organisms of the same species or different species, and also allows analysis of differences between a newly discovered protein sequence and the ones related to it.

In the specification, there are a number of examples which are proof of concept. Several of the examples are summarized here.

Prediction of anti-infective annotation in Mycobacterium tuberculosis

Seven outlier sequences were identified in *Mycobacterium tuberculosis* (Tables 1 & 2). Among these, three protein sequences corresponded to the glycine rich protein PE_PGRS (Poly E rich proteins) of *M. tuberculosis*. The amino acid sequences of these can be retrieved from the NCBI database. The PE_PGRS proteins have been implicated in virulence in this pathogen (Ramakrishnan et al 2000). These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

Prediction of anti-infective annotation in Helicobacter pylori

Eight outlier sequences were identified in *Helicobacter pylori* (Tables 1 & 2). One of these outliers is a histidine rich protein. The bacterial strain lacking the histidine rich protein is more susceptible to bismuth and Ni^{2+} than the wild-type strain (Mobley et al 19990. These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

Prediction of anti-infective annotation in Plasmodium falciparum

Five outlier sequences were identified in *Plasmodium falciparum* (Tables 1 & 2). The circumsporozite protein was evaluated as a vaccine candidate (Kester et al 2001). These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach .

Prediction of anti-infective annotation in Vibrio cholerae

Twelve outlier sequences were identified in *Vibrio cholerae* (Tables 1 & 2). One of these outlier proteins is to 1A. The 1A protein provides resistance to CTXϕ infection, which encodes cholera toxin (Heilpern et al 2000). These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

Prediction of anti-infective annotation in Mycoplasma genetalium

Four outlier sequences were identified in *Mycoplasma genetalium* (Tables 1 & 2). Two of these outlier proteins are cyto-adherence accessory proteins. In *Mycoplasma genetalium* a limited genetic capacity and lack of metabolic pathways is compensated by surface parasitism. And the close contact with the host cell is enhanced by the expression of cyto-adherence

accessory proteins (Wasinger et al, 2000). These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

Prediction of anti-infective annotation in Mycoplasma pneumoniae

Six outlier sequences were identified in *Mycoplasma pneumoniae* (Tables 1 & 2). One of these outlier proteins is an adhesin related protein. Adhesins and adhesin-related accessory proteins in *Mycoplasma pneumoniae* are required for cyto-adherence and the subsequent development of disease pathology (Reddy et al, 1995). This unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

Prediction of anti-infective annotation in Leishmania major

Ten outlier sequences were identified in *Leishmania major* (Tables 1 & 2). Two of these outlier proteins are hydrophilic surface protein. A family of differentially expressed genes from *Leishmania major* contains one sequence (Gene B) that encodes a novel, hydrophilic protein found on the surface of infective parasite stages (Flinn et al 1994). These unique outlier protein sequences can therefore be predicted to be potential candidates for an anti-infective approach.

References:

1. Chakravarti et al, *Vaccine* 19:601-612 (2000a)
2. Chakravarti et al, *Dev. Biol.* (Basel), 103:81-90 (2000b)
3. Flinn et al, *Mol. Biochem. Parasitol.*, 65(2):25970 (1994)
4. Fraser et al, *Emerging Infectious Diseases*, 6:505-512.38 (2000)
5. Guruprasad et al, *Protein Eng. Dec.*, 4(2):155-161 (1990)
6. Heilpern et al, *J. Bacteriol.*, 182(6):1739-1747 (2000)
7. Kester et al, *J. Infect. Dis.*, 183(4):640-647 (2001)
8. Liu et al, *Proc. Natl. Acad. Sci. USA*, 96(12):7011-7016 (1999).
9. Mande et al, *Biomol. Str. Dyn.*, 18:137-144 (2000)
10. Mobley et al, *Helicobacter*, 4(3):162-169 (1999)

11. Nakashima et al, *Mol. Biol.*, 238:54-61 (1994)

12. Nakashima et al, *J. Biochem. (Tokyo)*, 99(1):153-162 (1986)

13. Nakayama et al, *Chem. Inf. Comput. Sci.*, 28(2):72-78 (1988)

14. Nakashima et al, *FEBS Lett.*, 303(2-3):141-146 (1992)

15. Nandi et al, *J. Biosci.*, 27(1) Suppl. 1:15-25 (2002)

16. Ramakrishnan et al, A Novel Complexity Measure for Comparative Analysis of Protein Sequences from Complete, *J. Biomol. Str Dyn.*, (In press)

17. Natesh et al, *J. Mol. Biol.*, 288(5):999-1012 (1999)

18. Nishikawa et al, *J. Biochem.*, 94:997-1007 (1983)

19. Ramachandran et al, *Int. J. Pept. Proton Res.*, 31:1-16 (1988)

20. Ramakrishnan et al, *Science*, 288(5470):1436-1439 (2000)

21. Reddy et al, *J. Bacteriol.*, 177(20):5943-5951 (1995)

22. Schneider et al, *Gene Sep.* 3, 237(1):113-121 (1999)

23. Varadarajan et al, *Proc. Nacl. Acad.. Sci. USA*, 93:13908-13913 (1996)

24. Wasinger et al, *Eur. J. Biochem.*, 267(6):1571-1582 (2000).

In view of the discussion above, Applicants assert that a specific, substantial and credible utility has been established for the method recited in the pending claims. Therefore, Applicants respectfully request reconsideration and withdrawal of this rejection.

V. Rejection of the Claims Under 35 U.S.C. §112

A. At page 8 of the Office Action, last paragraph, the Examiner rejects claims 1-16 under 35 U.S.C. §112, first paragraph, as not being enabled.

The Examiner states that the claims do not define an use for the proteins identified in the claims. The Examiner further states that as the claims are allegedly not supported by a specific, substantial and credible utility, or a well-established utility, the skilled artisan would not know how to use the claimed invention.

In response, Applicants assert that, for the reasons discussed above, the claimed invention does have a patentable utility. Furthermore, the skilled artisan would clearly understand, based on the claims and the specification, how to use the claimed method in the identification of candidate anti-infective proteins. In addition, an use for the proteins is clear from the disclosure of the specification, namely in tests to determine whether the candidate protein will function as an anti-infective.

Accordingly, Applicants assert that the claimed invention is enabled, and therefore respectfully request reconsideration and withdrawal of this rejection.

B. At page 9 of the Office Action, second-to-last paragraph, the Examiner rejects claims 1-16 under 35 U.S.C. §112, second paragraph, as being indefinite.

Claim 1

The Examiner states that this claim is vague and indefinite because it is unclear how the claimed method involving protein outliers computationally validates the protein sequences as anti-infectives when no method step mentioned seems to support this declaration.

In response, Applicants first note that the claims do not recite a method of validating a protein sequence as an anti-infective. Instead, the method identifies proteins as candidate anti-infectives. Biochemical testing will be required to determine whether a candidate identified by the method may successfully be used as an anti-infective.

In addition, Applicants have amended the claim to clarify that step vi) is used to validate candidate anti-infectives.

In view of the amendment and this discussion, Applicants state that the claim is definite and therefore respectfully request reconsideration and withdrawal of this rejection.

Claims 2 and 9

The Examiner states that these claims contain embodiments which are beyond the elected invention. The Examiner proposes correcting the claims by deleting non-elected embodiments.

In response, Applicants note *B. burgdorferi* was elected "for search purposes" as stated in the Response to Election/Restriction Requirement filed in this application on October 17, 2002.

Furthermore, in the Office Action dated September 18, 2002, the Examiner states that "upon the allowance of a generic claim, applicant will be entitled to consideration of claims to additional species which are written in dependent form or otherwise include all the limitations of an allowed generic claim as provided in 37 C.F.R. §1.141."

While the Examiner has not yet allowed a generic claim, neither has the Examiner identified any prior art that reads on the elected species. Therefore, the scope of claims 2 and 9 is not improper at this stage of prosecution.

The Examiner also states that these claims are vague and indefinite due to the abbreviations used in the claims.

In response, Applicants note that in the amendment to the claims included herewith, the full name of the genus of each organism is now recited. In view of this amendment, Applicants state that the cited claims are definite as written, and therefore respectfully request reconsideration and withdrawal of this rejection.

Claims 11 and 14-16

The Examiner states that these claim recite the word "hypothetical" which is vague and indefinite.

In response, the term has been cancelled from the amended claims. Applicants therefore respectfully request reconsideration and withdrawal of this rejection.

Claim 13

The Examiner states that this claim is vague and indefinite due to the use of the abbreviation "CPU."

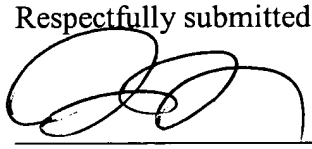
In response, the abbreviation has been spelled out in the amended claims. Applicants therefore respectfully request reconsideration and withdrawal of this rejection.

VI. Conclusion

In view of the above, reconsideration and allowance of this application are now believed to be in order, and such actions are hereby solicited. If any points remain in issue which the Examiner feels may be best resolved through a personal or telephone interview, the Examiner is kindly requested to contact the undersigned at the telephone number listed below.

The USPTO is directed and authorized to charge all required fees, except for the Issue Fee and the Publication Fee, to Deposit Account No. 19-4880. Please also credit any overpayments to said Deposit Account.

Respectfully submitted,



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